

XhoI hGH Signal Sequence

hGH Intron

TCGAG ATG GCT ACA G GTAAAGCGCCCTAAATCCCTTTGGGCACAAATGTGTCTGAGGCTAGAGGCAGCGACCTGTAGATGGGACGGGGGCACTAACCCCTCAGGTTTGGGGCTT
 ▶ Met Ala Thr

GAATGTGAGTATCGCCATGTAAGCCCATTTGGCCAATCTCAGAAAGCTCCTGGTCCCTGGAGGGATGGAGAGAGAAAAACAAACAGCTCCTGGAGCAGGGAGAGTGTGGCCTCTTGCTCTC

CGGCTCCCTCTGTTGCCCTCTGGTTTCTCCCCAGGC TCC CCG ACG TCC CTG CTC CTG GCT TTT GGC CTG CTC TGC CTG CCC TGG CTT

▶ Ser Arg Thr Ser Leu Leu Leu Ala Phe Gly Leu Leu Cys Leu Pro Trp Leu

+20 Asp of Processed TBP1

CAA GAG GGC AGT GGC GAT AGT GTG TGT CCC CAA GGA AAA TAT ATC CAC CCT CAA AAT AAT TCG ATT TGC TGT ACC AAG TGC CAC AAA GGA
 ▶ Gln Glu Gly Ser Ala Asp Ser Val Cys Pro Gln Gly Lys Tyr Ile His Pro Gln Asn Asn Ser Ile Cys Cys Thr Lys Cys His Lys Gly

ACC TAC TTG TAC AAT GAC TGT CCA GGC CCG GGG CAG GAT ACG GAC TGC AGG GAG TGT GAG AGC GGC TCC TTC ACC GCT TCA GAA AAC CAC CTC
 ▶ Thr Tyr Leu Tyr Asn Asp Cys Pro Gly Pro Gly Gln Asp Thr Asp Cys Arg Glu Cys Glu Ser Gly Ser Phe Thr Ala Ser Glu Asn His Leu

AGA CAC TGC CTC AGC TGC TCC AAA TGC CGA AAG GAA ATG GGT CAG GTG GAG ATC TCT TCT TGC ACA GTG GAC CGG GAC ACC GTG TGT GGC TGC
 ▶ Arg His Cys Leu Ser Cys Ser Lys Cys Arg Lys Glu Met Gly Gln Val Glu Ile Ser Ser Cys Thr Val Asp Arg Asp Thr Val Cys Gly Cys

ACG AAG AAC CAG TAC CGS CAT TAT TGG AGT GAA AAC CTT TTC CAG TGC TTC AAT TGC AGC CTC TGC CTC AAT GGG ACC GTG CAC CTC TCC TGC
 ▶ Arg Lys Asn Gln Tyr Arg His Tyr Trp Ser Glu Asn Leu Phe Gln Cys Phe Asn Cys Ser Leu Cys Leu Asn Gly Thr Val His Leu Ser Cys

CAG GAG AAA CAG AAC ACC GTG TGC ACC TGC CAT GCA GGT TTC TTT CTA AGA GAA AAC CAG TGT GTC TCC TGT GGC GGT GCT GGC CCA GGT
 ▶ Gln Glu Lys Gln Asn Thr Val Cys Thr Cys His Ala Gly Phe Phe Leu Arg Glu Asn Glu Cys Val Ser Cys Ala Gly Ala Ala Pro Gly

TGC CCA GAA TGC ACG CTA CAG GAA AAC CCA TTC TTC TCC CAG CCG GGT GCC CCA ATA CTT CAG TGC ATG GGC TGC TGC TTC TCT AGA GCA TAT
 ▶ Cys Pro Glu Cys Thr Leu Gln Glu Asn Pro Phe Phe Ser Gln Pro Gly Ala Pro Ile Leu Gln Cys Met Gly Cys Cys Phe Ser Arg Ala Tyr

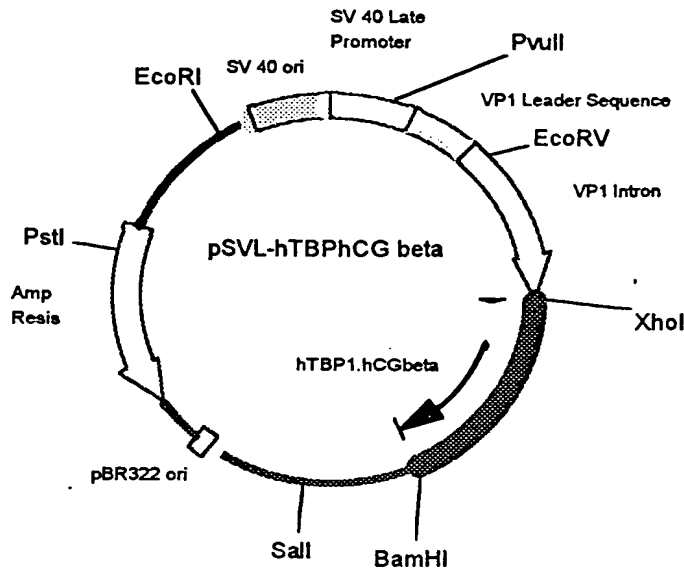
CCC ACT CCA CTA AGG TCC AAG AAG ACG ATG TTG GTC CAA AAG AAC GTC ACC TCA GAG TCC ACT TGC TGT GTA GCT AAA TCA TAT AAC AGG GTC
 ▶ Pro Thr Pro Leu Arg Ser Lys Lys Thr Met Leu Val Gln Lys Asn Val Thr Ser Glu Ser Thr Cys Cys Val Ala Lys Ser Tyr Asn Arg Val

ACA GTA ATG GGG GGT TTC AAA GTG GAG AAC CAC ACG GCG TGC CAC TGC AGT ACT TGT TAT TAT CAC AAA TCT TAA G
 ▶ Thr Val Met Gly Gly Phe Lys Val Glu Asn His Thr Ala Cys His Cys Ser Thr Cys Tyr Tyr His Lys Ser ...

BamHI

Figure 1 (a)

TBP(20-161)-hCG α FUSION CONSTRUCT



hGH Signal Sequence

hGH Intron

XhoI

CTCGAG ATG GCT ACA G GTAAGCGCCCTAAATCCCTTTGGGCACAAATGTGTCTGAGGGGAGAGGCGACCTGTAGATGGGACGGGGGCACTAACCTCAGGTTTGGG
 ▶ Met Ala Thr

GCTTCTGAATGTGAGTATCGCCATGTAAGCCAGTATTTGGCCAAATCTCGAAAGCTCCTGGTCCCTGGAGGGATGGGAGAGTAAACAAACAGCTCCTGGAGCGGGAGAGTCTGTGGC

CTCTTGCTCTCCGGCTCCCTCTGTTGCCCTCTGGTTTCTCCCCAGGC TCC CGG ACC TCC CTG CTC CTG GCT TTT GGC CTG CTC TGC CTG
 ▶ Ser Arg Thr Ser Leu Leu Leu Ala Phe Gly Leu Leu Cys Leu
 +20 Asp of Processed TBP1

CCC TGG CTT CAA GAG GGC AGT GCC GAT AGT GTG TGT CCC CAA GGA AAA TAT ATC CAC CCT CAA AAT AAT TCG ATT TGC TGT ACC
 ▶ Pro Trp Leu Gln Glu Gly Ser Ala Asp Ser Val Cys Pro Gln Gly Lys Tyr Ile His Pro Gln Asn Asn Ser Ile Cys Cys Thr

AAG TGC CAC AAA GGA ACC TAC TTG TAC AAT GAC TGT CCA GGC CCG GGG CAG GAT ACG GAC TGC AGG GAG TGT GAG AGC GGC TCC TTC ACC
 ▶ Lys Cys His Lys Gly Thr Tyr Leu Tyr Asn Asp Cys Pro Gly Pro Gly Gln Asp Thr Asp Cys Arg Glu Cys Glu Ser Gly Ser Phe Thr

GCT TCA GAA AAC CAC CTC AGA CAC TGC CTC AGC TGC TCC AAA TGC CGA AAG GAA ATG GGT CAG GTG GAG ATC TCT TCT TGC ACA GTG GAC
 ▶ Ala Ser Glu Asn His Leu Arg His Cys Leu Ser Cys Ser Lys Cys Arg Lys Glu Met Gly Gln Val Glu Ile Ser Ser Cys Thr Val Asp

CGG GAC ACC GTG TGT GGC TGC AGG AAG AAC CAG TAC CGG CAT TAT TGG AGT GAA AAC CTT TTC CAG TGC TTC AAT TGC AGC CTC TGC CTC
 ▶ Arg Asp Thr Val Cys Gly Cys Arg Lys Asn Gln Tyr Arg His Tyr Trp Ser Glu Asn Leu Phe Gln Cys Phe Asn Cys Ser Leu Cys Leu

AAT GGG ACC GTG CAC CTC TCC TGC CAG GAG AAA CAG AAC ACC GTG TGC ACC TGC CAT GCA GGT TTC TTT CTA AGA GAA AAC GAG TGT GTC
 ▶ Asn Gly Thr Val His Leu Ser Cys Gln Glu Lys Gln Asn Thr Val Cys Thr Cys His Ala Gly Phe Phe Leu Arg Glu Asn Glu Cys Val

TCC TGT GCT GGT GCT GGT CCA CGG TGC CGC CCC ATC AAT GCC ACC CTG GCT GTG GAG AAG GAG GGC TGC CCC GTG TGC ATC ACC GTC
 ▶ Ser Cys Ala Gly Ala Gly Pro Arg Cys Arg Pro Ile Asn Ala Thr Leu Ala Val Glu Lys Glu Gly Cys Pro Val Cys Ile Thr Val

AAC ACC ACC ATC TGT GCC GGC TAC TGC CCC ACC ATG ACC CGC GTG CTG CAG GGG GTC CTG CCG GCC CTG CCT CAG GTG GTG TGC AAC TAC
 ▶ Asn Thr Thr Ile Cys Ala Gly Tyr Cys Pro Thr Met Thr Arg Val Leu Gln Gly Val Leu Pro Ala Leu Pro Gln Val Val Cys Asn Tyr

CGC GAT GTG CGC TTC GAG TCC ATC CGG CTC CCT GGC TGC CCG CGC GGC GTG AAC CCC GTG GTC TCC TAC GCC GTG GCT CTC AGC TGT CAA
 ▶ Arg Asp Val Arg Phe Glu Ser Ile Arg Leu Pro Gly Cys Pro Arg Gly Val Asn Pro Val Val Ser Tyr Ala Val Ala Leu Ser Cys Gln

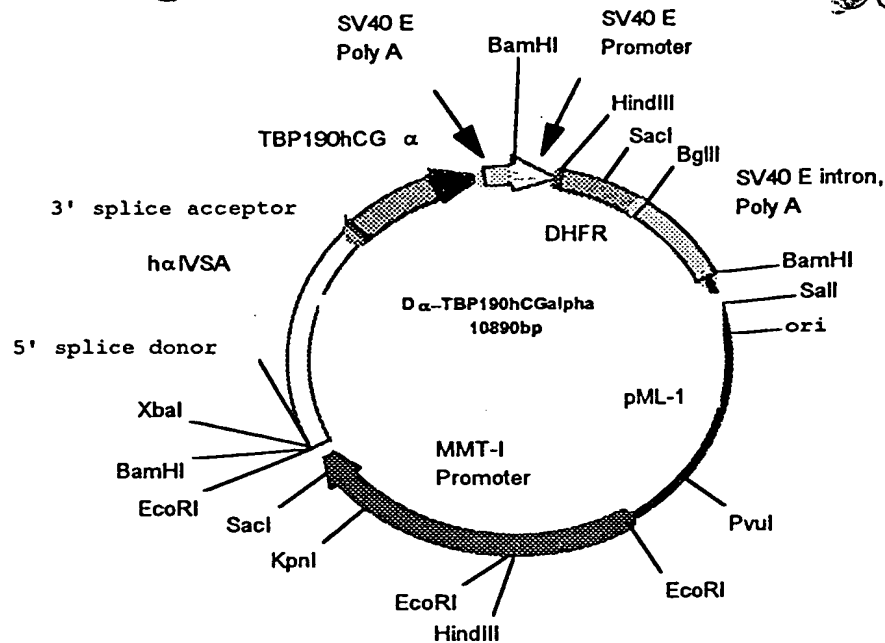
TGT GCA CTC TGC CGC CGC AGC ACC ACT GAC TGC GGG GGT CCC AAG GAC CAC CCC TTG ACC TGT GAT GAC CCC CGC TTC CAG GAC TCC TCT
 ▶ Cys Ala Leu Cys Arg Arg Ser Thr Thr Asp Cys Gly Gly Pro Lys Asp His Pro Leu Thr Cys Asp Asp Pro Arg Phe Gln Asp Ser Ser

TCC TCA AAG GCC CCT CCC CCC AGC CTT CCA AGC CCA TCC CGA CTC CCG GGG CCC TCG GAC ACC CCG ATC CTC CCA CAA TAA

▶ Ser Ser Lys Ala Pro Pro Pro Ser Leu Pro Ser Pro Ser Arg Leu Pro Gly Pro Ser Asp Thr Pro Ile Leu Pro Gln ... Bam HI

Figure 1 (b)

TBP(20-161)-hCGβ FUSION CONSTRUCT



XhoI hGH Signal Sequence hGH Intron

TCGAG ATG GCT ACA G GTAAGCGCCCTAAATCCCTTTGGGCACAATGTGTCTGAGGGGAGAGGCAGCGACCTGTAGATGGGACGGGGGCACTAACCCCTCAGGTTTGGGGCTTCT

► Met Ala Thr

GAATGTGAGTATCGCCATGTAAGCCCACTATTGGCCCAATCTCAGAAAAGCTCCTGGTCCCTGGAGGGATGGAGAGAGAAAAACAAACAGCTCCTGGAGCAGGGAGAGTGTGGCCCTCTTGTCTCTC

CGGCTCCCTCTGTTGCCCTCTGGTTTCTCCCCAGGC TCC CGG ACG TCC CTG CTC CTG GCT TTT GGC CTG CTC TGC CTG CCC TGG CTT

► Ser Arg Thr Ser Leu Leu Leu Ala Phe Gly Leu Leu Cys Leu Pro Trp Leu

+20 Asp of processed TBP1

CAA GAG GGC AGT GCC GAT AGT GTG TGT CCC CAA GGA AAA TAT ATC CAC CCT CAA AAT AAT TCG ATT TGC TGT ACC AAG TGC CAC AAA GGA

► Gln Glu Gly Ser Ala Asp Ser Val Cys Pro Gln Gly Lys Tyr Ile His Pro Gln Asn Asn Ser Ile Cys Cys Thr Lys Cys His Lys Gly

ACC TAC TTG TAC AAT GAC TGT CCA GGC CCG GGG CAG GAT ACG GAC TGC AGG GAG TGT GAG AGC GGC TCC TTC ACC GCT TCA GAA AAC CAC CTC

► Thr Tyr Leu Tyr Asn Asp Cys Pro Gly Pro Gly Gln Asp Thr Asp Cys Arg Glu Cys Glu Ser Gly Ser Phe Thr Ala Ser Glu Asn His Leu

AGA CAC TGC CTC AGC TGC TCC AAA TGC CGA AAG GAA ATG GGT CAG GTG GAG ATC TCT TCT TGC ACA GTG GAC CGG GAC ACC GTG TGT GGC TGC

► Arg His Cys Leu Ser Cys Ser Lys Cys Arg Lys Glu Met Gly Gln Val Glu Ile Ser Ser Cys Thr Val Asp Arg Asp Thr Val Cys Gly Cys

AGG AAG AAC CAG TAC CGG CAT TAT TGG AGT GAA AAC CTT TTC CAG TGC TTC AAT TGC AGC CTC TGC CTC AAT GGG ACC GTG CAC CTC TCC TGC

► Arg Lys Asn Gln Tyr Arg His Tyr Trp Ser Glu Asn Leu Phe Gln Cys Phe Asn Cys Ser Leu Cys Leu Asn Gly Thr Val His Leu Ser Cys

CAG GAG AAA CAG AAC ACC GTG TGC ACC TGC CAT GCA GGT TTC TTT CTA AGA GAA AAC GAG TGT GTC TCC TGT AGT AAC TGT AAG AAA AGC CTG

► Gln Glu Lys Gln Asn Thr Val Cys Thr Cys His Ala Gly Phe Phe Leu Arg Glu Asn Glu Cys Val Ser Cys Ser Asn Cys Lys Lys Ser Leu

GAG TGC ACG AAG TTG TGC CTA CCC CAG ATT GAG AAT GTT AAG GGC ACT GAG GAC TCA GGC ACC ACA GGC GGT GCT GGC CCA GGT TGC CCA

► Glu Cys Thr Lys Leu Cys Leu Pro Gln Ile Glu Asn Val Lys Gly Thr Glu Asp Ser Gly Thr Thr Ala Gly Ala Ala Pro Gly Cys Pro

GAA TGC ACG CTA CAG GAA AAC CCA TTC TTC TCC CAG CCG GGT GCC CCA ATA CTT CAG TGC ATG GGC TGC TGC TTC TCT AGA GCA TAT CCC ACT

► Glu Cys Thr Leu Gln Glu Asn Pro Phe Phe Ser Gln Pro Gly Ala Pro Ile Leu Gln Cys Met Gly Cys Cys Phe Ser Arg Ala Tyr Pro Thr

CCA CTA AGG TCC AAG AAG ACG ATG TTG GTC CAA AAG AAC GTC ACC TCA GAG TCC ACT TGC TGT GTA GCT AAA TCA TAT AAC AGG GTC ACA GTA

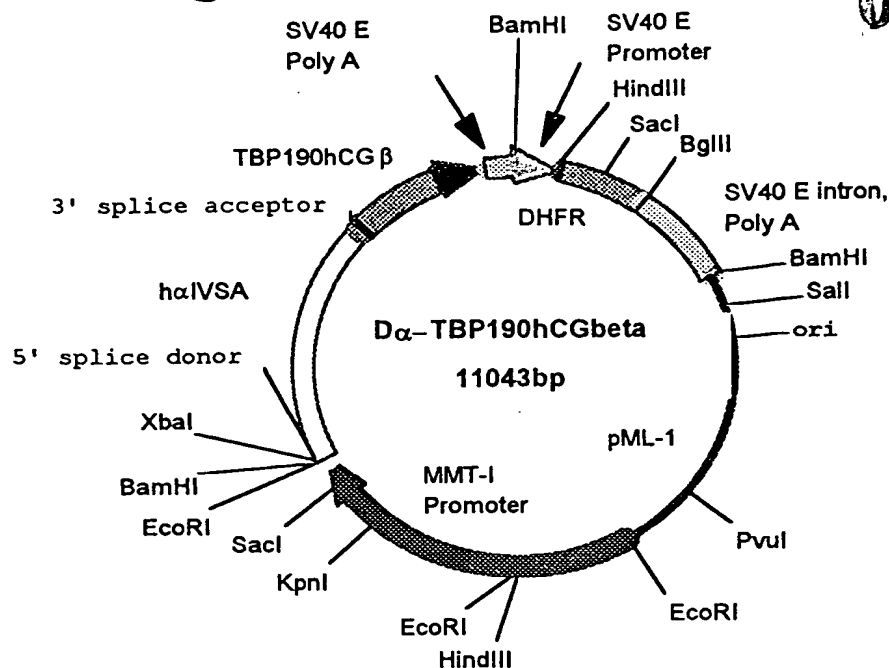
► Pro Leu Arg Ser Lys Lys Thr Met Leu Val Gln Lys Asn Val Thr Ser Glu Ser Thr Cys Cys Val Ala Lys Ser Tyr Asn Arg Val Thr Val

ATG GGG GGT TTC AAA GTG GAG AAC CAC ACG GCG TGC CAC TGC AGT ACT TGT TAT TAT CAC AAA TCT TAA GGATCCCTCGAG

► Met Gly Gly Phe Lys Val Glu Asn His Thr Ala Cys His Cys Ser Thr Cys Tyr Tyr His Lys Ser ... BamHI XhoI

Linker +7 Cys of hCG alpha

Figure 2(a)
TBP(20-190)-hCG α FUSION CONSTRUCT



XhoI hGH Signal Sequence hGH Intron
 CTCGAG ATG GCT ACA G GTAAAGCGCCCTAAATCCCTTTGGGCAATGTCTCCTGAGGGGAGAGGCGACCTTAGATGGGACGGGGGCACTAACCTCAGGTTTGGG
 ▶Met Ala Thr

GCTTCTGAATGTGAGTATCGCCATGTAGCCCGAGTATTTGGCCCAATCTCAGAAAGCTCCTGGTCCCTGGAGGGATGGAGAGAGAAAAACAAACAGCTCCTCGGACAGGGAGAGTGTCTGGC

CTCTTGTCTCCGGCTCCCTCTGTTCGCCCTCTGGTTTCCTCCCGAGGC TCC CGG ACG TCC CTG CTC CTG GCT TTT GGC CTG CTC TGC CTG
 ▶Ser Arg Thr Ser Leu Leu Leu Ala Phe Gly Leu Leu Cys Leu

+20 Asp of Processed TBP1

CCC TGG CTT CAA GAG GGC AGT GGC GAT AGT GTG TGT CCC CAA GGA AAA TAT ATC CAC CCT CAA AAT AAT TCG ATT TGC TGT ACC
 ▶Pro Trp Leu Gln Glu Gly Ser Ala Asp Ser Val Cys Pro Gln Gly Lys Tyr Ile His Pro Gln Asn Asn Ser Ile Cys Cys Thr

AAG TGC CAC AAA GGA ACC TAC TTG TAC AAT GAC TGT CCA GGC CCG GGG CAG GAT ACG GAC TGC AGG GAG TGT GAG AGC GGC TCC TTC ACC
 ▶Lys Cys His Lys Gly Thr Tyr Leu Tyr Asn Asp Cys Pro Gly Pro Gly Gln Asp Thr Asp Cys Arg Glu Cys Glu Ser Gly Ser Phe Thr

GCT TCA GAA AAC CAC CTC AGA CAC TGC CTC AGC TGC TCC AAA TGC CGA AAG GAA ATG GGT CAG GTG GAG ATC TCT TCT TGC ACA GTG GAC
 ▶Ala Ser Glu Asn His Leu Arg His Cys Leu Ser Cys Ser Lys Cys Arg Lys Glu Met Gly Gln Val Glu Ile Ser Ser Cys Thr Val Asp

CGG GAC ACC GTG TGT GGC TGC AGG AAG AAC CAG TAC CGG CAT TAT TGG AGT GAA AAC CTT TTC CAG TGC TTC AAT TGC AGC CTC TGC CTC
 ▶Arg Asp Thr Val Cys Gly Cys Arg Lys Asn Gln Tyr Arg His Tyr Trp Ser Glu Asn Leu Phe Gln Cys Phe Asn Cys Ser Leu Cys Leu

AAT GGG ACC GTG CAC CTC TCC TGC CAG GAG AAA CAG AAC ACC GTG TGC ACC TGC CAT GCA GGT TTC TTT CTA AGA GAA AAC GAG TGT GTC
 ▶Asn Gly Thr Val His Leu Ser Cys Gln Glu Lys Gln Asn Thr Val Cys Thr Cys His Ala Gly Phe Phe Leu Arg Glu Asn Glu Cys Val

TCC TGT AGT AAC TGT AAG AAA AGC CTG GAG TGC ACG AAG TTG TGC CTA CCC CAG ATT GAG AAT GTT AAG GGC ACT GAG GAC TCA GGC ACC
 ▶Ser Cys Ser Asn Cys Lys Lys Ser Leu Glu Cys Thr Lys Leu Cys Leu Pro Gln Ile Glu Asn Val Lys Gly Thr Glu Asp Ser Gly Thr

Linker +7 Pro of beta
 ACA GCT GGT GCT GGT CCA CGG TGC CGC CCC ATC AAT GCC ACC CTG GCT GTG GAG AAG GAG GGC TGC CCC GTG TGC ATC ACC GTC AAC
 ▶Thr Ala Gly Ala Gly Pro Arg Cys Arg Pro Ile Asn Ala Thr Leu Ala Val Glu Lys Glu Gly Cys Pro Val Cys Ile Thr Val Asn

ACC ACC ATC TGT GCC GGC TAC TGC CCC ACC ATG ACC CGC GTG CTG CAG GGG GTC CTG CCG GCC CTG CCT CAG GTG GTG TGC AAC TAC CGC
 ▶Thr Thr Ile Cys Ala Gly Tyr Cys Pro Thr Met Thr Arg Val Leu Gln Gly Val Leu Pro Ala Leu Pro Gln Val Val Cys Asn Tyr Arg

GAT GTG CGC TTC GAG TCC ATC CGG CTC CCT GGC TGC CGC GGC GTG AAC CCC GTG GTC TCC TAC GCC GTG GCT CTC AGC TGT CAA TGT
 ▶Asp Val Arg Phe Glu Ser Ile Arg Leu Pro Gly Cys Pro Arg Gly Val Asn Pro Val Val Ser Tyr Ala Val Ala Leu Ser Cys Gln Cys

GCA CTC TGC CGC CGC AGC ACC ACT GAC TGC GGG GGT CCC AAG GAC CAC CCC TTG ACC TGT GAT GAC CCC CGC TTC CAG GAC TCC TCT TCC
 ▶Ala Leu Cys Arg Arg Ser Thr Thr Asp Cys Gly Gly Pro Lys Asp His Pro Leu Thr Cys Asp Asp Pro Arg Phe Gln Asp Ser Ser Ser

TCA AAG GCC CCT CCC CCC AGC CTT CCA AGC CCA TCC CGA CTC CCG GGG CCC TCG GAC ACC CCG ATC CTC CCA CAA TAA GGATCCCTCGAG
 ▶Ser Lys Ala Pro Pro Pro Ser Leu Pro Ser Pro Ser Arg Leu Pro Gly Pro Ser Asp Thr Pro Ile Leu Pro Gln ... BamHI XhoI

Figure 2 (b)
 TBP(20-190)-hCGβ FUSION CONSTRUCT

Figure 4. CHO cell expressed TBP-hCG(20-190) inhibits TNF α -induced cytotoxicity on BT-20 cells

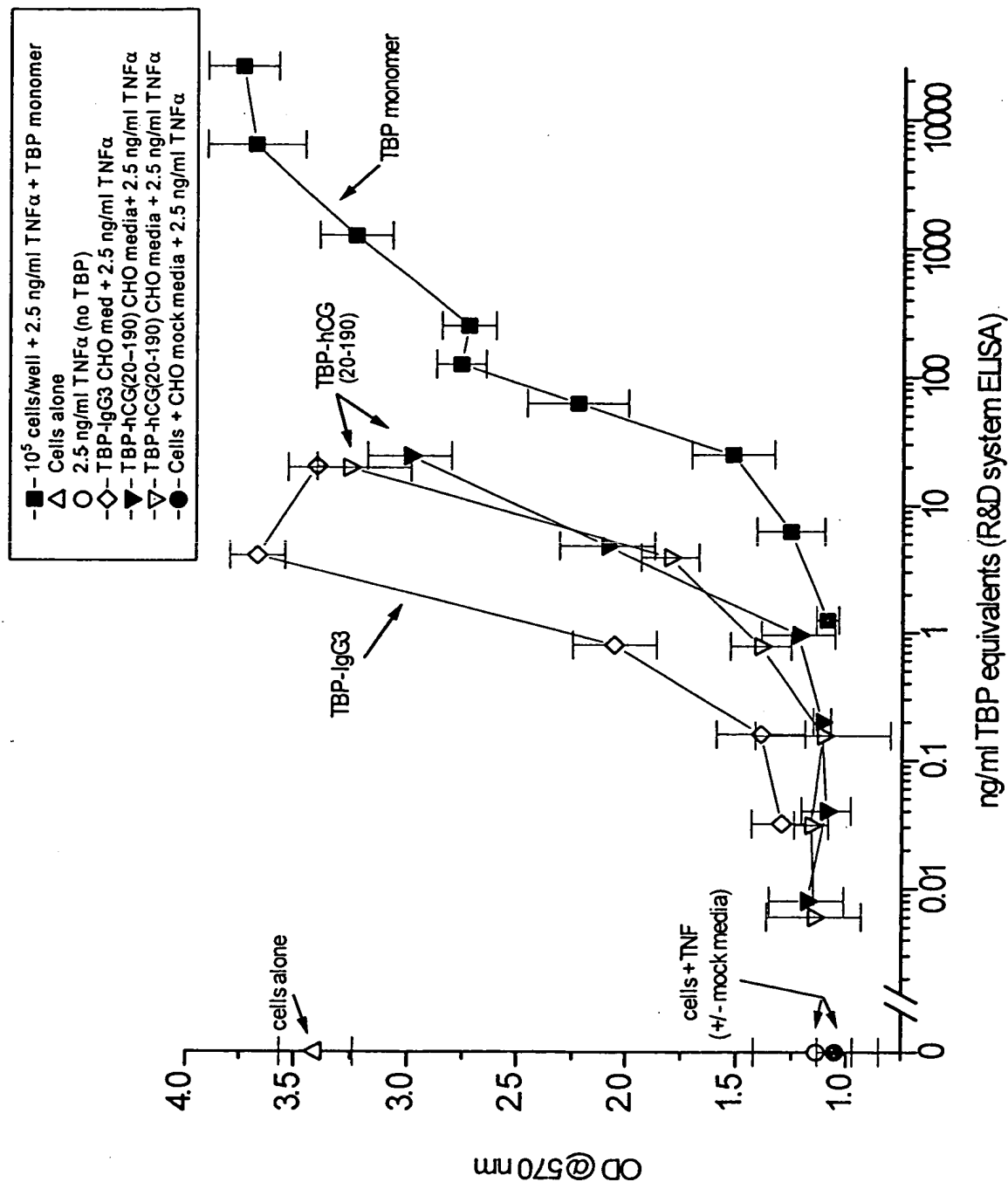


Figure 5. COS cell expressed TBP-hCG(20-190) inhibits TNF α -induced cytotoxicity on BT-20 cells

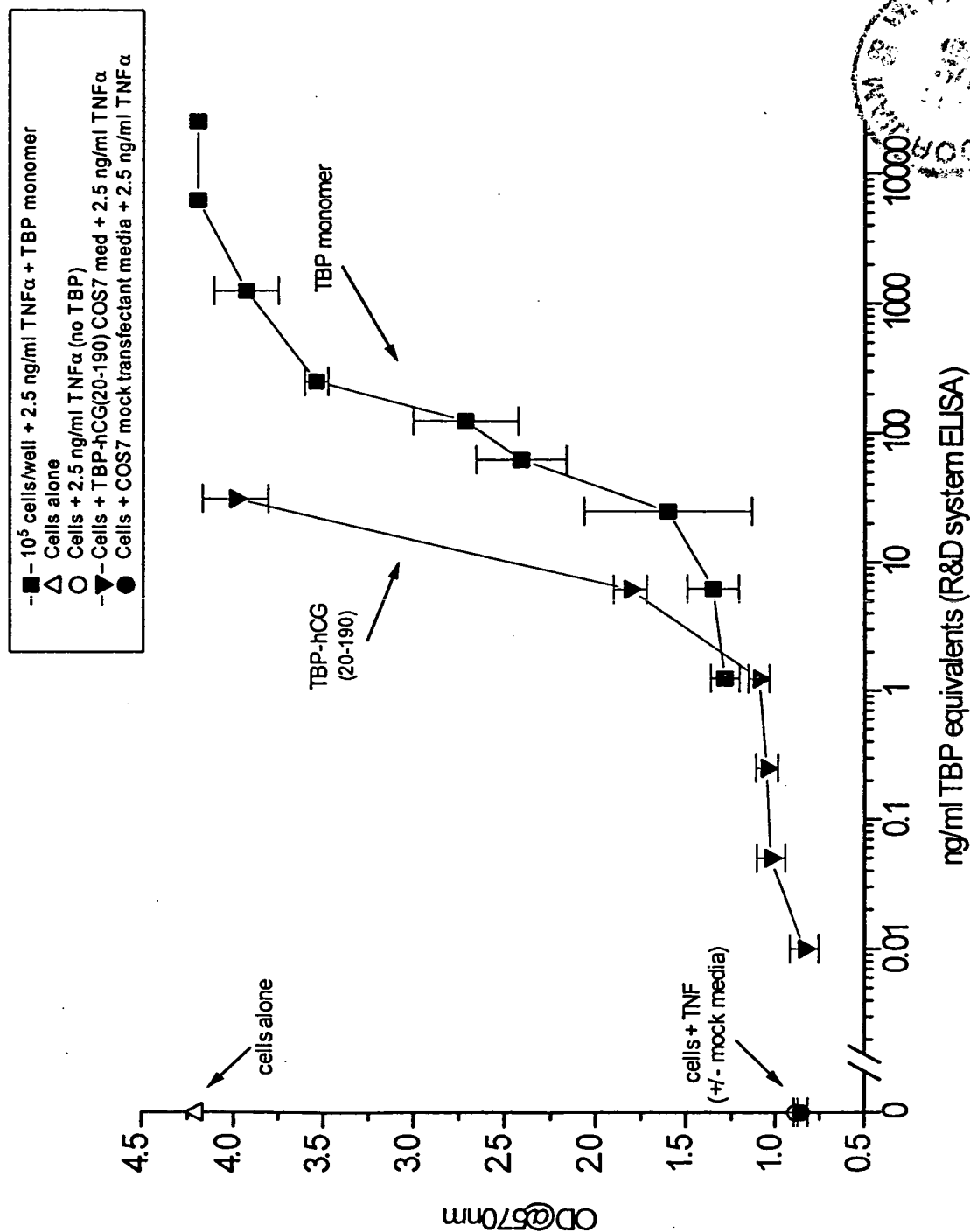


Figure 6. Affinity purified CHO cell expressed TBP-hCG(20-161) inhibits TNF α -induced cytotoxicity on BT-20 cells

